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Applying Navy Dolphin Medicine to Conservation Medicine for Small Cetaceans

Anwendung der Delfinmedizin der Navy auf die Artenschutzmedizin für kleine Cetaceen

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Abstract

Marine mammal veterinarians play an important role in the conservation of small cetaceans, specifically with regard to the application of their unique expertise to wild animal health assessments and the continual advancement of medical tools, techniques, and approaches to cetacean care. The US Navy Marine Mammal Program (Navy) has cared for common bottlenose dolphins (*Tursiops truncatus*) for over 60 years and has established a comprehensive clinical practice aimed at providing the best possible care to Navy animals, per US Secretary of the Navy instruction. To date, the Navy has contributed more than 1200 publications to marine mammal science, ranging from discoveries in physiology, acoustics, and anatomy, to advanced diagnostic techniques for marine mammal healthcare. This knowledge has been directly applied to the conservation of at-risk, threatened, and endangered small cetaceans, especially dolphins. Three case studies demonstrate the translation of Navy medicine to conservation medicine, to include: (1) conducting diagnostic ultrasound evaluations of wild dolphins following the *Deepwater Horizon* oil spill; (2) predicting health impacts of freshwater exposure on wild dolphins; and (3) developing minimally-invasive aging techniques for dolphins. The case examples are relevant to all veterinarians caring for and/or evaluating the health of small cetaceans in zoo-

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logical settings, marine mammals rehabilitation centers, or in the wild. The aquatic mammal medical community must continue to find opportunities to translate managed animal medicine and care into conservation medicine, integrating our veterinary knowledge into conservation action plans for at-risk cetaceans and species recovery.

Keywords: dolphin, veterinary, medicine, conservation, cetaceans

Introduction

The US Navy Marine Mammal Program (Navy) has cared for common bottlenose dolphins (*Tursiops truncatus*) for more than 60 years (NIWC Pacific, 2023). Navy dolphins spend a portion of their time swimming freely in the open ocean as part of operational training plans and in support of the animals' health and welfare. When the animals are not at sea, they are cared for in open ocean enclosures, which are located in bays, sounds, and estuaries co-inhabited by local marine life. The US Secretary of the Navy dictates that Navy marine mammals are provided the highest quality of humane care and treatment (SECNAV INSTRUCTION 3900.41H, 2018). Caring for the animals in this way has allowed veterinarians, animal care experts, and biologists to continuously monitor their health and behavior in a controlled, natural environment that closely mimics the living conditions of their wild counterparts (Houser, Finneran, & Ridgway, 2010; Smith et al., 2012; Smith et al., 2013a; Venn-Watson, Jensen, Smith, Xitco, & Ridgway, 2015; Linnehan et al., 2020; Ivančić et al., 2020). The high quality care provided in an ocean environment has resulted in long average lifespans, with Navy dolphins living an average of 32.5 years (Venn-Watson et al., 2015).

The Navy is a leader in marine mammal science and medicine, contributing more than 1200 publications to the scientific literature, ranging from discoveries regarding basic physiologic, acoustic, and anatomic adaptations to the ocean environment, to advanced diagnostic techniques for marine mammal healthcare (Department of the Navy, 1998; Ridgway, 2008; Houser et al., 2010; NIWC Pacific, 2023). Since 2007, the National Marine Mammal Foundation (NMMF) has provided medical, training, and research support to the Navy. To date, the NMMF has added 350 publications to the marine mammal peer-reviewed literature, many of which involve Navy animals (NMMF, 2023). Much of the knowledge gained can be directly applied to the conservation of at-risk small cetaceans. This has led to a rich history of translating Navy medicine into conservation tools, techniques, and strategies for wild marine mammals, facilitated through partnerships with nonprofit organizations, government agencies, expert consultants, private institutions, and universities (Houser et al., 2010; NIWC Pacific, 2023; NMMF, 2023).

Over the years, direct applications have included conservation planning for endangered cetaceans such as the Yangtze river dolphin (*Lipotes vexillifer*) in China (Ridgway, Norris, & Cornell, 1998) and the vaquita porpoise (*Phocoena sinus*) in Mexico (Rojas-Bracho et al., 2019); medical database development for the Southern Resident killer whale (*Orcinus orca*) in the US Pacific Northwest (Lutmerding et al., 2019); advancement of diagnostic techniques for application to field assessments of small cetaceans (Smith et al., 2013a; Wells et al., 2014; Ivančić et al., 2020; Linnehan et al., 2020; Barratclough et al., 2021a); health assessments of at-risk cetaceans including those impacted by the *Deepwater Horizon* (DWH) oil spill in the northern Gulf of Mexico (Schwacke et al., 2014; Smith et al., 2017; Barratclough et al., 2019a; Linnehan et al., 2021; Smith et al., 2022); and the development of minimally-invasive aging techniques to inform population assessments of marine mammals (Barratclough et al., 2019b; Barratclough et al., 2021b; Barratclough et al., 2023). Through improvements in care provided to the Navy's marine mammals, we are developing and applying tools, techniques, and knowledge gained to the conservation of at-risk marine mammals.

To demonstrate the translation of Navy dolphin medicine to conservation medicine, we will review the following three case examples where Navy medical knowledge was applied to wild cetacean conservation efforts: (1) conducting diagnostic ultrasound evaluations of wild dolphins following the DWH oil spill; (2) predicting health impacts of freshwater exposure on dolphins; and (3) developing minimally-invasive aging techniques for dolphins.

Case One: Applying Diagnostic Ultrasound Techniques to Wild Dolphin Health Assessments Following the *Deepwater Horizon* Oil Spill

The DWH oil spill resulted in large-scale contamination of bays, sounds, and estuaries in the northern Gulf of Mexico (Michel et al., 2013). More than 1000 miles of shoreline were oiled along the US Gulf Coast from Texas to Florida (ERMA, 2015). NOAA conducted a Natural Resource Damage Assessment (NRDA) to determine the potential adverse health effects of the oil spill and associated contaminants on wildlife (DWH NRDA Trustees, 2016). Catch-and-release health assessments of wild dolphins were performed in Barataria Bay, Louisiana, and Mississippi Sound, MS, which were heavily impacted by the oil spill (Schwacke et al., 2014; Smith et al., 2017). Marine mammal veterinarians from diverse backgrounds were called upon to apply their collective expertise to the damage assessment. More than 150 live bottlenose dolphins were examined during the catch-and-release health assessments from 2011 to 2014. These comprehensive medical exams utilized numerous diagnostic tools and techniques developed with marine mammals in human care, summarized in Schwacke et al. (2014) and Smith et al. (2017).

Diagnostic ultrasound was an essential tool used in the wild dolphin medical exams, providing a rapid and data-rich evaluation of internal organ health. Full body ultrasounds were conducted by experienced sonographers (CRS, FMG, & JMM) either in water, out of the water, or a combination of both, on all dolphins stable enough for the procedure (Fig. 1). Portable, rugged ultrasound units outfitted with heads-up displays were utilized in the field, based on proven use with Navy dolphins and other managed marine mammals in various ocean environments (Fig. 2). Ultrasound protocols developed for and validated with Navy dolphins were employed in the field, including evaluation of lungs, marginal lymph nodes, kidneys, liver, pancreas, and when applicable, fetal health (Venn-Watson, Smith, Johnson, Daniels, & Townsend, 2010; Smith et al., 2012; Smith et al., 2013a; Smith et al., 2013b; Seitz, Smith, Marks, Venn-Watson & Ivančić, 2016; Martony et al., 2017; Ivančić et al., 2020). Additional protocols developed by aquarium and zoologic institutions were utilized to evaluate the dolphin gastrointestinal system, ovaries, testes, and fetal growth (Williamson et al., 1990; Robeck et al., 1998; Brook et al., 2000; Brook, 2001; Lacave et al., 2004; Robeck et al., 2005; Yuen et al., 2009; Fiorucci et al., 2015; Saviano et al., 2020). Decades of sonographer experience with dolphins in human care were applied to the determination of normal versus abnormal organ health.

Pulmonary ultrasound evaluation proved to be particularly important in this case, aiding in the documentation of an increased prevalence of moderate to severe lung disease in dolphins living within the oil spill footprint (Barataria Bay, LA, and Mississippi Sound, MS) versus those living in an unoiled bay (Sarasota Bay, FL) (Schwacke et al., 2014). Although there was some evidence that pulmonary health was slightly improving four years after the spill (Smith et al., 2017), ultrasound examinations performed in 2017 and 2018 showed persistent and even worsening pulmonary disease in oil-impacted dolphins (Smith et al., 2022). Based

Fig. 1: A diagnostic ultrasound exam of a wild dolphin during NOAA's DWH oil spill investigation performed by an experienced sonographer (CRS). The animal was placed into a stretcher and then transferred to the deck of a vessel for examination. The sonographer used a portable ultrasound unit outfitted with heads-up-display goggles and wore a dark visor to block out the sun, allowing for better visualization of the projected image. The animal was monitored by veterinary and animal care staff, and a portable ECG unit outfitted with 'sticky' leads was used to monitor the heart rate. (Photo: NOAA, Permit #932-1905/MA-009526)

Fig. 2: A routine diagnostic ultrasound procedure with a Navy dolphin performed by an experienced sonographer (FMG). A portable ultrasound unit was utilized, outfitted with heads-up-display goggles. The animal voluntarily slid into an animal transporter and was carefully transferred to the deck for examination. She was continuously monitored by veterinary and animal care staff, and her heart rate was monitored with a portable ECG unit outfitted with suction cup electrodes. (Photo: US Navy)

on the comprehensive health data available for these dolphins, chronic pulmonary disease was likely a significant factor in the overall poor health of dolphins living within the DWH oil spill footprint, defined in Schwacke et al. (2022). As determined by diagnostic ultrasound, dolphins born *after* the DWH oil spill did not have an elevated prevalence of pulmonary disease. Additional catch-and-release studies are needed to determine if the prevalence of lung disease in dolphins born after the spill remains low, and if dolphins alive during the DWH spill event show evidence of improvement, stabilization, or further degradation over time (Schwacke et al., 2023).

In addition to lung disease, veterinarians utilized other diagnostic tests to diagnose numerous adverse health effects in live dolphins impacted by the DWH oil spill, which included an impaired stress response, altered immune response, poor body condition, and increased mortality rates (Schwacke et al., 2014; DWH NRDA Trustees, 2016; Smith et al., 2017; Takeshita et al., 2017; de Guise et al., 2021). At the conclusion of NOAA's damage assessment, questions remained regarding the long-term impacts of oil exposure. For example, dolphins living within the oil spill footprint were experiencing high rates of reproductive failure, but the underlying etiology was unknown. Additionally, there were concerns regarding the potential for an emerging cardiac issue in dolphins, which had been documented in fish, birds, and humans impacted by the oil spill (reviewed in Takeshita et al, 2021a).

In order to better understand the reproductive and cardiac health effects in wild dolphins, we needed more advanced ultrasound techniques. We carefully examined our Navy medicine needs and looked for overlap and intersections between what would benefit Navy dolphins and wild dolphins. As a result, we began work with Navy dolphins to refine procedures for reproductive ultrasound and echocardiography, and then applied the advanced reproductive and cardiac health techniques to wild dolphins impacted by the oil spill. This led to novel protocols, based on human fetal ultrasound techniques, that allowed for the rapid assessment of fetal, placental, and maternal health in pregnant Barataria Bay dolphins (Ivančić et al., 2020), as well as the ability to comprehensively evaluate the health of adult dolphin hearts (Linnehan et al., 2020; Linnehan et al., 2021). At the conclusion of the study, we had diagnosed placental dysfunction in the majority of Barataria Bay dolphin pregnancies, and we detected cardiac anomalies in several of the adult dolphins living within the oil spill footprint. Overall, the Navy dolphin medical program allowed for the advancement of diagnostic imaging techniques to improve Navy animal care, the care of other dolphins in human care, and the rapid assessment of at-risk dolphins in the wild.

Case Two: Predicting Health Impacts of Freshwater Exposure on Wild Dolphins

In the aftermath of the DWH oil spill disaster, a major restoration project has been planned for Barataria Bay, Louisiana, called the Mid-Barataria Sediment Diversion (MBSD) project (Louisiana Deepwater Horizon Trustee Implementation Group, 2021; U.S. Army Corps of Engineers, 2023). The MBSD project is meant to partially restore marsh in the Barataria basin and reduce land loss along the barrier islands. Central to the plan is the controlled diversion of the Mississippi River into Barataria Bay, which would carry nutrients and sediment into the basin to help rebuild wetlands. According to the US Army Corp of Engineers' Final Environmental Impact Statement (2023), the MBSD project would create a maximum of 27 square miles of land build in the mid-Barataria Basin by 2050. In the process, the salinity of the bay would be intermittently and substantially reduced, which could cause significant harm to Barataria Bay dolphins. These dolphins have demonstrated strong site fidelity and are unlikely to leave despite low salinity conditions (Lane et al., 2015; Wells et al., 2017; Takeshita et al., 2021b; Schwacke et al., 2022; Thomas et al., 2022).

Numerous questions have emerged during consideration of the planned restoration project, including the following: (1) how long can bottlenose dolphins survive in a freshwater and/or a very low salinity environment; (2) what sub-clinical and clinical physiologic effects occur and on what timeline in low saline environments; and (3) when do freshwater-induced skin lesions begin to occur? Navy clinicians began data-mining historical records to look for relevant data to help address these conservation questions, since operational Navy dolphins have traveled to various salinity environments throughout their lifetime. Over several decades, data was collected and archived from the animals in various aquatic environments. These data were collated and analyzed to help characterize physiologic responses to low salinity and to develop a timeline of potential health consequences (McClain et al., 2020). The study documented changes in serum electrolytes, osmolality, adrenal hormones, and skin integrity in response to low salinity conditions.

The Navy dolphin dataset provided unique and otherwise difficult to obtain information about bottlenose dolphin physiology and health effects of a specific environmental challenge. The study was made available through peer-review publication to conservationists, environmental planners, and policy makers, and informed an expert elicitation and subsequent modeling effort aimed at predicting how the MBSD project would impact Barataria Bay dolphins (Schwacke et al. 2022; Thomas et al. 2022). The resulting model predicted that the ~2000 bottlenose dolphins living in Barataria Bay are unlikely to survive prolonged periods of freshwater. If the MBSD project moves forward as proposed, Barataria Bay dolphins could go functionally extinct within 50 years (Thomas et al., 2022). This case example demonstrates how archived Navy medical data can help inform scientific predictions regarding the potential consequences of habitat alteration and environmental changes to an at-risk dolphin population.

Case Three: Developing Minimally-Invasive Aging Techniques for Wild Dolphins

Estimating the age of individual animals in wild dolphin populations, especially those at-risk, is essential to understanding population demographics, interpreting biological data, and determining recovery trajectories (Schwacke et al., 2021). For decades, the gold standard of age estimation in live and dead bottlenose dolphins involved counting growth layer group (GLG) in a longitudinal section of a mandibular tooth, requiring tooth extraction and analysis (Ridgway et al., 1975; Hohn, 1980; Hohn et al., 1989; Read et al., 1993). Some variation has been documented in GLG-based age estimates (Kimura, 1980; Barratclough et al, 2023); however, it was the best available option for decades. During live animal catch-and-release health assessments, veterinary dental and anesthetic techniques were refined to allow for the safe and humane extraction of a single tooth for this purpose (reviewed in Townsend, Smith, & Rowles, 2018). In an effort to replace invasive techniques with less invasive options, marine mammal veterinarians led the development of minimally-invasive methods for age estimation. These alternative methods included the radiographic assessment of pectoral flipper bone maturation, dental X-ray analysis, and epigenetic age assessment.

Pectoral flipper radiography technique development for age estimation of bottlenose dolphins was led by D. García-Párraga in an aquarium setting, with collaboration from multiple institutions caring for bottlenose dolphins, including the Navy (Barratclough et al., 2019b). Pectoral flipper radiographs were performed with dolphins of known ages and changes in bone maturation were assigned numerical scores in order to calculate age estimates. Ages could be properly estimated with decreasing precision as animals aged; specifically, within 3 months in dolphins < 5 years old, to within 5 years in dolphins > 30 years old. The technique was applied by Barratclough et al. (2023) to wild bottlenose dolphins and Navy dolphins of known ages for comparison to dental GLG analysis. Pectoral flipper radiograph age estimates were comparable in accuracy to dental GLG analysis, establishing the usefulness of this minimally-invasive radiographic procedure for age estimation in live bottlenose dolphins. The technique allowed for same-day age estimates across a broad range of age categories. It should be noted that a dental X-ray technique for age estimation was also established with wild dolphins of known ages using the pulp:tooth area ratio method (Herrman, 2020). This method can reliably provide same-day estimates for bottlenose dolphin ages up to about 10 years of age.

Epigenetic age estimation based on DNA methylation was refined for managed bottlenose dolphins using archived Navy dolphin blood and skin samples (Barratclough et al., 2021b). DNA methylation is the addition of methyl groups to cytosine-phosphate-guanine (CpG) sites and occurs systematically with age, therefore methylation assessment can provide a chronological age estimate. Epigenetic age analysis has been further evaluated for wild bottlenose dolphins with a focus on skin samples collected via dart biopsy, in order to provide a remote option for age estimates of free-ranging animals. Additionally, epigenetic aging methods have been applied to an at-risk population of bottlenose dolphins, Lahille's dolphins (*Tursiops truncatus gephyreus*) found along the east coast of South America (Barratclough et al., 2022). Approximately 600 Lahille's dolphins remain and only 360 are considered capable of reproducing (Vermeulen et al., 2019). Providing age-related context for existing biological data and population demographics could enhance species recovery plans.

The managed animal community as a whole contributed to these important technique advancements for estimating dolphin ages using minimally-invasive techniques, which will likely replace the more invasive tooth extraction procedure in wild dolphins. The Navy dolphin population played an important role by assisting with the validation of radiographic and epigenetic techniques, specifically through the utilization of archived biological samples from known age animals collected periodically over the animals' lifetime. Collaborations among veterinarians caring for dolphins in zoological settings and rehabilitation centers allowed for rapid progress in the development of minimally invasive techniques, effectively advancing marine mammal medicine for the benefit of both managed and wild dolphins.

Conclusion

Marine mammals in human care have a clear role in the conservation of at-risk species, specifically with regard to advancements of medical tools, diagnostic techniques, and veterinary approaches. Caring for managed marine mammals provides invaluable comparisons to wild animals, including physiologic and pathologic responses to environmental challenges. Veterinary practitioners charged with caring for marine mammals have unique skillsets that enhance our ability to rapidly, safely, and accurately examine and assess wild marine mammals. Navy clinicians who are charged with providing the highest quality care to the Navy's dolphins have demonstrated the effective application of their medical practice to the conservation of at-risk cetaceans. The examples provided are relevant to all veterinarians caring for small cetaceans in zoological settings and marine mammal rehabilitation centers, and during health assessments of wild animals. The aquatic mammal medical community must continue to translate managed animal medicine into conservation medicine. With aquatic mammals around the world facing continued anthropogenic threats and ever-changing environmental stressors, integrating veterinary knowledge into conservation action plans for species recovery and survival is critical.

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Zusammenfassung

Tierärzte, die auf marine Säugetiere spezialisiert sind, spielen eine wichtige Rolle bei dem Schutz von Kleinwalen, insbesondere im Hinblick auf die Anwendung ihres Fachwissens bei der Bewertung des Gesundheitszustands von Wildtieren und die kontinuierliche Weiterentwicklung medizinischer Instrumente, Techniken und Ansätze für die Pflege dieser Tiere. Das Programm für Meeressäuger der US-Marine (Navy) hält seit über 60 Jahren Große Tümmler (Tursiops truncatus) und hat eine umfassende klinische Praxis etabliert, die darauf abzielt, auf Anweisung des US-Marineministers die bestmögliche Pflege für Navy-Tiere zu gewährleisten. Bis heute hat die Marine mit mehr als 1200 Veröffentlichungen zur Forschung an Meeressäugern beigetragen. Die Studien reichen von Entdeckungen in der Physiologie, der Akustik und Anatomie bis hin zu fortschrittlichen Diagnosetechniken für die medizinische Versorgung von Meeressäugern. Dieses Wissen wurde direkt für den Schutz gefährdeter, bedrohter und vom Aussterben bedrohter Kleinwale, insbesondere von Delfinen, eingesetzt. In drei Fallstudien wird gezeigt, inwieweit Wissen aus der Veterinärmedizin des Navy Programms für den Schutz von Delphinarten angewendet werden kann, darunter: (1) Durchführung von diagnostischen Ultraschalluntersuchungen bei wild lebenden Delfinen nach der Deepwater Horizon Ölkatastrophe; (2) Vorhersage der gesundheitlichen Auswirkungen von Süßwasser auf wild lebende Delfine; und (3) Entwicklung minimal-invasiver Methoden zur Altersbestimmung von Delfinen. Die Fallbeispiele sind für alle Tierärzte relevant, die sich in zoologischen Einrichtungen, Rehabilitationszentren für Meeressäuger oder in freier Wildbahn um Kleinwale kümmern und/oder deren Gesundheit beurteilen. Die Gemeinschaft der auf Meeressäuger spezialisierten Tiermediziner sollte weiterhin an Möglichkeiten arbeiten um ihr Fachwissen in Aktionspläne für den Schutz gefährdete Delphine zu integrieren.

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